

## The chromosomes of *Erebia ligea rishirizana* (Nymphalidae, Satyrinae)

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### Summary

The chromosome number of *Erebia ligea rishirizana* males from Hokkaido, Japan is 56 in diploid (brain cells and spermatogonia) and 28 in haploid (primary and secondary spermatocytes), while 29 chromosomes have been counted in the first division of *E. ligea* females from Finland (Federley, 1938).

### Zusammenfassung

Die Chromosomenzahl bei *Erebia ligea rishirizana* Männchen von Hokkaido, Japan, ist 56 in diploidem (Zellen des ZNS und Spermatogonien) und 28 in haploidem Zustand (Spermatocyten I. und II. Ordnung), während 29 Chromosomen in der ersten Reifeteilung bei *E. ligea* Weibchen aus Finnland ermittelt worden waren (Federley, 1938).

### Résumé

Le nombre de chromosomes chez des mâles de *Erebia ligea rishirizana*, en provenance de Hokkaido au Japon, est de 56 à l'état diploïde (cellules du cerveau et spermatogonies) et de 28 à l'état haploïde (spermatocytes de premier et de deuxième ordre), alors que 29 chromosomes ont été comptés en première division chez des femelles de *E. ligea* de Finlande (Federley, 1938).

The genus *Erebia* (Nymphalidae Satyrinae) comprises a good many taxa karyologically examined. Previously, Federley (1938) reported the occurrence of 29 bivalents in females of *Erebia ligea* (Linnaeus, 1758) from Finland, though no chromosome figures of them are shown. As well known, this species ranges widely from Europe to the Far Eastern regions of Asia. In Japan, two subspecies are found : *E. ligea rishirizana* Matsumura, 1928 in Hokkaido and *E. ligea takanonis* Matsumura, 1909 in Central Honshû. Recently, we have had the opportunity to

examine karyologically the former subspecies from Japan. The results of this chromosome examination are described below.

### Material and methods

Adult females caught on a woodland path through Muri-dake (Mt. Muri) of Hokkaido, Japan laid eggs. Laboratory-reared pupae and larvae were exclusively used for the present chromosome examination.

Both brains and testes were taken out from all males, and the brains from females too. All of the brains and some of the testes were fixed in Carnoy (3 : 1). These were carefully acid-dissociated and air-dried. Chromosome spreads of these preparations were stained with Giemsa (4 %). Other testes were squashed with lacto-acetic orcein for the examination of spermatocyte chromosomes.

### Observations and remarks

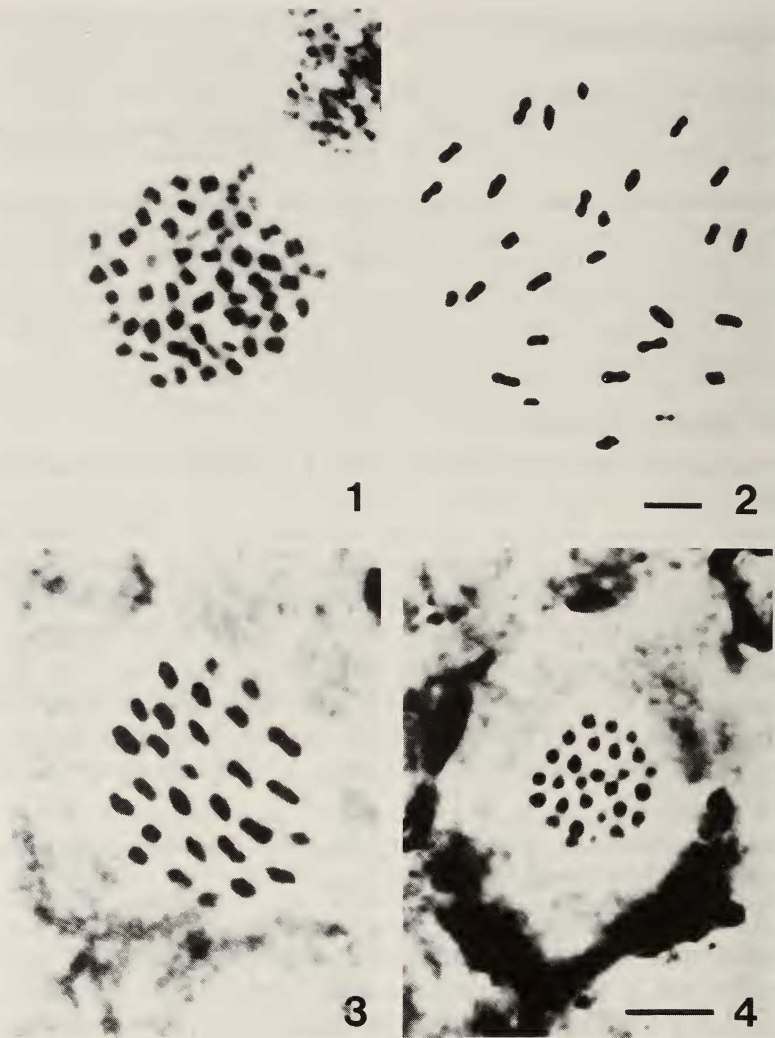
Chromosomes were successfully observed in males. Both diploid and haploid chromosome numbers, 56 and 28, were determined with certainty in the acid-dissociated preparations (figs 1, 2); counts were made in metaphases of thirteen brain cells and thirty spermatogonia, and in those of twenty-six primary spermatocytes and twenty-four secondary spermatocytes from five mature final instar larvae.

The haploid chromosome number, 28, was confirmed also with testis-squashes (figs 3, 4); counts were made in metaphases of 409 primary spermatocytes and 118 secondary spermatocytes from three pupae and three young final instar larvae. Variation in the haploid number was not observed.

From these findings, it is safe to conclude that the males of this subspecies have a  $2n, 56$ ;  $n, 28$  karyotype.

In fig. 1, a spermatogonial metaphase ( $2n, 56$ ) is shown as a representative of the diploid complements examined. The haploid complements in the first and second divisions ( $n, 28$ ) are shown in figs 2, 3, 4. As evident from these, the haploid chromosomes are round, or somewhat oval in shape in polar view. The chromosomes which are remarkable in behaviour are not observed in the present material.

It has thus become evident that *E. ligea* from Finland ( $n, 29$ ) and *E. ligea rishirizana* ( $n, 28$ ) examined here differ from each other in their chromosome constitution. Therefore, a comparison of their chromosome complements is indispensable for phyloanalysis of *E. ligea* and its subspecies. Consequently, a karyological re-examination of *E.*



Figs 1-4. Diploid and haploid germ-line chromosomes of *Erebia ligea rishirizana* males. 1 & 2. Chromosome complements from acid-dissociated testes (Giemsa): 1 — Spermatogonial mitosis ( $2n, 56$ ); 2 — First division ( $n, 28$ ). Scale bar: ca. 5  $\mu$ . 3 & 4. Haploid chromosome complements from testis-squashes (Orcein): 3 — First division ( $n, 28$ ); 4 — Second division ( $n, 28$ ). Scale bar: ca. 5  $\mu$ .

*ligea* from Finland is necessary, because, as stated above, mention was made of its haploid number alone (Federley, 1938).

On one hand, another subspecies in Japan, *E. ligea takanonis* still remains unexplored karyologically. We are intending to examine its chromosomes in the nearest future.

The congener in Japan, *E. nipponica* Janson, 1877 has a quite distinct karyotype of  $2n, 38$ ;  $n, 19$  (male) (Saitoh, 1988; 1989; Saitoh *et al.*, 1991a, b). Therefore, the inquiry into the chromosome complement is one of the reliable ways to discern these two Japanese taxa of *E. ligea rishirizana* and *E. nipponica*.

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